



Analysis of Antiasthmatic Drugs by HPLC

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Abstract

Theophylline is a bronchodilator used in treatment of acute and chronic asthma in newborn children. Because of its small therapeutic index, narrow therapeutic range and significant interpatient variability in pharmacokinetics its concentration has to be monitored closely. Another antiasthmatic drug, caffeine has some advantages over theophylline, for example, more potency to stimulate the central nervous system, a wide therapeutic index and a long half-life. Different HPLC, GC and immunoassays are available for drug level measurement of antiasthmatic drugs.

The HPLC method presented here shows the chromatogram of the four common antiasthmatic drugs theophylline, theobromine, caffeine and emprophylline using isocratic analysis on a reversed phase column and UV detection. To avoid decomposition of samples the autosampler temperature was set to 4 °C.

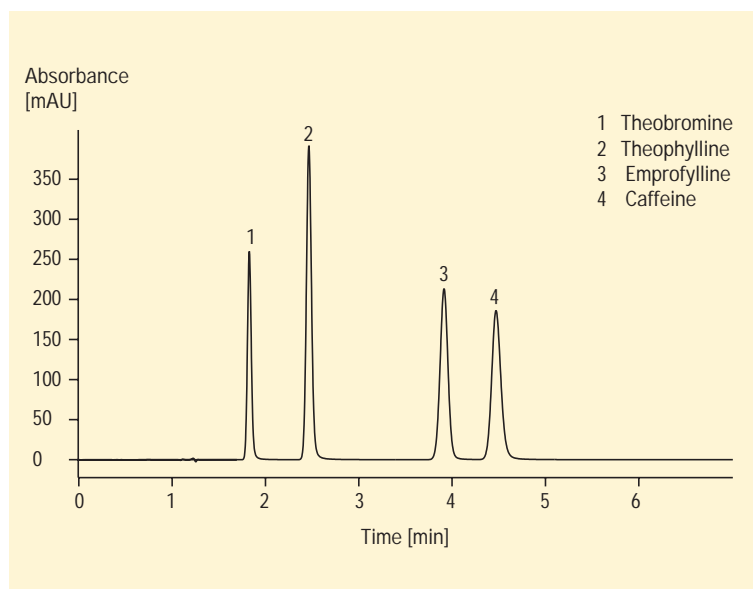


Figure 1
Analysis of four antiasthmatic drugs

Conditions

Column

4 x 125 mm ODS Hypersil 5 µm

Mobile phase

A = water

B = acetonitrile

Flow rate 1.0 ml/min

Isocratic 8 % B for 7 min

UV detector

diode array detector 270/20 nm,
reference 360/80 nm standard cell
or variable wavelength detector
270 nm, standard cell

Column compartment temperature
50 °C

Stop time 7 min

Injection volume 5 µl



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The performance of the HPLC method is shown in the table below.

Compound	LOD for S/N=3 (mg/l)*	Precision of RT (RSD of 10 runs) (100 mg/l)*	Precision of Area (RSD of 10 runs) (100 mg/l)*
Theobromine	0.1	0.09	0.12
Theophylline	0.1	0.09	0.19
Emprofylline	0.1	0.09	0.24
Caffeine	0.1	0.10	0.08

* Injection volume: 5 µl

The method presented here shows an easy but reliable and precise analysis of the antiasthmatic drugs theophylline, theobromine, caffeine and emprofylline. The values for LOD, precision of RT and precision of area show the good performance of the analysis

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Equipment

Agilent 1100 Series

- Quaternary pump (includes vacuum degasser)
- Thermostatted autosampler
- Thermostatted column compartment
- Variable wavelength detector, standard flow cell, 10-mm path length, 13-µl cell volume

Alternative:

- Vacuum degasser
- Binary pump
- Diode-array detector, standard flow cell 10-mm path length, 13-µl cell volume
- Agilent ChemStation + 3D software

Columns

- Hypersil ODS, 5 µm, 4 x 125 mm (Agilent part number 792618-564)
- *Recommended:* Guard cartridges Hypersil ODS, 5 µm, 4 x 4 mm (Agilent part number 7992618-504, 10/pk)

Note:

Since the method was specifically developed on the Agilent 1100 Series system you might not be able to reproduce this analysis on an older system or even on a new system with lower performance. To avoid sample decomposition it is necessary to use a cooled autosampler, for example, the Agilent 1100 Series thermostatted autosampler.



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